Index

MONWEALTH INST.
OMOLOGY LIBRARY

Aus. HO

Vol. 9, No. 4. December, 1952.

THE QUEENSLAND JOURNAL OF AGRICULTURAL SCIENCE

CONTENTS

PAGE.

Effects of Methyl-Thiouracil on Liver Vitamin A, Spermatogenesis and Calcium Metabolism of Merino Rams. By W. T. K. HALL and J. M. HARVEY .. 301

The Oxalate Content of Some Queensland Plants. By R. H. MATHAMS and A. K. SUTHERLAND 317

Three New Species of Dacinae (Trypetidae, Diptera) from Queensland. By A. W. S. MAY 335

PUBLISHED QUARTERLY

By the Queensland Department of Agriculture and Stock, Brisbane

Issued by Direction of Hon H. H. COLLINS,
Secretary for Agriculture and Stock.

EDITORIAL BOARD.

Chairman and Editor: C. W. WINDERS, B.Sc.Agr., A.C.I.S.

A. L. CLAY, B.V.Sc., Assistant Director, Division of Animal Industry;

J. LEGG, B.Sc., D.V.Sc., M.R.C.V.S., Director of Research, Division of Animal Industry;

E. B. RICE, Dip.Ind.Chem., Director of Dairying, Division of Dairying;

J. H. SIMMONDS, M.B.E., M.Sc., Officer in Charge, Science Branch, Division of Plant Industry;

W. J. S. SLOAN, M.Sc.Agr., Assistant Director of Agriculture, Agriculture Branch, Division of Plant Industry;

J. H. SMITH, M.Sc., N.D.A., Assistant Director of Horticulture, Horticulture Branch, Division of Plant Industry;

A. K. SUTHERLAND, B.V.Sc., M.S., Senior Veterinary Pathologist, Division of Animal Industry;

L. G. VALLANCE, M.Sc., Assistant Director, Bureau of Sugar Experiment Stations;

M. WHITE, M.Sc., Ph.D., A.R.A.C.I., Agricultural Chemist and Biochemist, Chemical Laboratory, Division of Plant Industry.

The Journal is supplied free of charge to organizations and institutions engaged in agricultural science or veterinary science. Applications should be made to the Under Secretary, Department of Agriculture and Stock, Brisbane, Queensland, Australia.

THE QUEENSLAND JOURNAL OF AGRICULTURAL SCIENCE

VOL. 9 - - NO. 4.
DECEMBER, 1952.

EFFECTS OF METHYL-THIOURACIL ON LIVER VITAMIN A, SPERMATOGENESIS AND CALCIUM METABOLISM OF MERINO RAMS.

By W. T. K. HALL, B.V.Sc. (Assistant Veterinary Officer, Animal Health Station, Yeerongpilly) and J. M. HARVEY, M.Sc. (Senior Chemist, Biochemical Section, Chemical Laboratory, Division of Plant Industry).

SUMMARY.

Four groups each of six Merino rams aged 1½-2 years at the beginning of the experiment were fed and treated as follows:—

Group I—Basal ration of bleached oaten chaff and linseed meal.

Group II—Basal ration plus 4-methyl-thiouracil.

Group III—Basal ration plus carotene.

Group IV—Basal ration plus 4-methyl-thiouracil plus carotene.

The dose rate of methyl-thiouracil was approximately 2·5 g. per 100 lb. body weight per day given by mouth.

Semen from all animals was examined every three weeks.

After approximately 100 days of treatment the animals were slaughtered, and thyroid weights, testes weights, serum calcium levels and liver vitamin A concentrations were determined.

The mean weight of the thyroids (16·5 g.) in Groups II and IV was over four times that of the thyroids in Groups I and III.

Serum calcium levels of rams in Groups II and IV were mainly lower (range 4·0–10·6 mg. per 100 ml.) than those of rams in Groups I and III (7·2–11·0 mg. per 100 ml.). The manner in which calcium metabolism was affected, either by the changed activity of the thyroid glands or as a side effect of the methyl-thiouracil treatment, is discussed.

Although there was wide variation in liver vitamin A concentration within the groups (as wide as 55 to 190 micrograms per gram), no differences between groups could be detected.

The lethargic clinical state of the animals and the grossly enlarged thyroids indicated that methyl-thiouracil had produced a hypothyroid state. It was concluded that the hypothyroid condition did not affect the storage of vitamin A in the liver. The hypothyroidism was more severe than that likely to be brought about in the field by starvation and/or by high atmospheric temperature.

No marked differences were apparent in the semen from the different groups.

A record is presented of the very low liver vitamin A concentration of 2 micrograms per gram for a ram which was given a greater aggregate amount of methyl-thiouracil than the other rams but which had been withdrawn from the experiment at an early stage because of small food intake, anaemia and jaundice.

INTRODUCTION.

Temporary infertility in sheep has been observed in north-western Queensland during the hot part of the year and particularly during dry years. Moule (1950) suggested that the so-called "summer sterility" in rams is one of the main causes. In these areas factors that may impair the fertility of rams are low intakes of dietary carotene, protein and phosphate and high atmospheric temperature (Gunn, 1936; Gunn, Sanders and Granger, 1942; Sapsford, 1951).

The thyroid activity of rats was shown by Dempsey and Astwood (1943) to be affected by high temperatures. It was reported by Eveleth, Bolin and Goldsby (1949) that thiouracil treatment of sheep appeared to prevent the conversion of carotene to vitamin A, and Canadell and Valdecases (1947) and Johnson and Baumann (1947) reported the same effect in rats. Best and Taylor (1945) stated: ". . . in hypothyroidism the conversion of carotene to the vitamin is defective; the milk of goats, which ordinarily is pure white, becomes yellow after thyroidectomy, due to the excretion of unchanged carotene."

It has therefore been suggested that in north-western Queensland hot weather depresses the activity of the thyroid and thus reduces the efficiency of conversion of the small amount of available carotene. This could produce a vitamin A deficiency, which in turn may affect the thyroid and so set up an adverse cycle (Moule, 1950). The experiment reported here was designed to test this hypothesis. Facilities for simulating the temperatures of north-western Queensland were not available, so methyl-thiouracil was used to depress the activity of the thyroid.

EXPERIMENTAL.

The objectives were to determine whether depressed thyroid activity affected the conversion of carotene to vitamin A, and whether vitamin A deficiency and hypothyroidism affected spermatogenesis in rams.

Twenty-seven 1½-2-year-old Merino rams were used. They were bred in southern Queensland and were brought to Yeerongpilly on June 8, 1950. To deplete their vitamin A reserves they were allowed no green feed and were fed the lightest coloured oaten hay procurable. During this period of depletion (on July 31) the rams were divided into three groups of seven and one group of six so that an animal of similar weight was in each group.

To provide information for comparison with other rams after treatment, one animal for post-mortem examination was selected at random from each of the three groups of seven animals. Thyroid, testis and epididymis were examined histologically, and liver vitamin A estimations were carried out.

The four groups, each of six animals, were then treated as follows during the experimental period, which began on November 17, 1950:—

Group I—Basal ration.

Group II—Basal ration plus methyl-thiouracil.

Group III—Basal ration plus carotene after the 8th day of the experiment (Nov. 25, 1950).

Group IV—Basal ration plus methyl-thiouracil then plus carotene after the 8th day of experiment.

The basal ration consisted of:—

Bleached oaten chaff ranging from 4·5 per cent. to 7·5 per cent. crude protein fed *ad lib.*

Linseed meal (minimum crude protein 30 per cent.), 4 oz. per sheep per day.

After the 51st day of the experiment ground limestone was mixed with the linseed meal at the rate of 2 oz. per six sheep per week.

METHODS.

General.

The sheep were fed twice a day. The amount of feed taken was about 2½ lb. per sheep per day, the methyl-thiouracil treated sheep eating slightly less than those not given methyl-thiouracil.

At three-weekly intervals the animals were weighed and plasma vitamin A and carotene estimations and semen evaluations were done. Red blood cell counts and serum calcium estimations were made as the clinical states of the animals indicated.*

* Clinical hypocalcaemia in the methyl-thiouracil treated groups was not expected, and consequently serum calcium estimations were not done until late in the experiment.

At autopsy, thyroid and testes were weighed and the liver vitamin A was estimated. Liver, thyroid, testis and epididymis were examined histologically.

Methyl-thiouracil Treatment.

Bogart and Mayer (1946) administered 10 g. thiouracil per day by mouth to produce "summer sterility" in rams. Astwood and Van der Laan (1945) stated that 6-methyl-thiouracil is five times more active in humans than thiouracil. As the drug available to us—4-methyl-thiouracil (British Drug Houses)—was assumed to have the same physiological activity as 6-methyl-thiouracil, a dose of 2 g. 4-methyl-thiouracil per day was used initially.

Since Ely, Olson and Reineke (1948) had shown that thiouracil was almost completely eliminated from the blood stream of calves and goats within 24 hours, the experimental animals were dosed at 9 a.m. and 5 p.m. each day. Dosing was by drenching, because methyl-thiouracil is not palatable to animals.

The initial dose of 2 g. of methyl-thiouracil per day for rams 90-110 lb. was changed after one week because the treated animals had not shown any loss of appetite or any other clinical change. The afternoon dose was increased by 50 per cent. The daily dose rates for the remaining 111 days of the experiment were then:—

Rams 70-90 lb. weight—2·0 g.
Rams 90-110 lb. weight—2·5 g.
Rams 110-130 lb. weight—3·0 g.

During the first three days the drug was given in gelatin capsules, but throughout the rest of the experiment it was administered as a drench containing 1 g. methyl-thiouracil per 5 ml. This drench was a paraffin suspension of finely ground methyl-thiouracil.

Dosing was discontinued or varied at times for clinical reasons as indicated later in the clinical reports.

Carotene Treatment.

Pure beta-carotene was not given until after eight days of methyl-thiouracil treatment in order to give the methyl-thiouracil time to act upon the thyroid. Carotene was then added to the protein supplement each week at the rate of 250 mg. per six sheep per week. This was done by thoroughly mixing into the linseed meal 0·25 g. of pure beta-carotene in 50 ml. of peanut oil containing 0.1 per cent. hydroquinone. The amount of carotene supplement per animal per day was based on the recommendation of the National Research Council, U.S.A. (1944).

Semen Evaluation.

Semen was collected by the electrical stimulation method used by Gunn (1936), and evaluated on volume, density, motility, pH drift and morphology.

Histological Examinations.

Liver and thyroid were fixed in 10 per cent. formalin saline, and testis and epididymis in Bouin's fluid for 18-20 hours. The tissues were stained with haemotoxylin and eosin.

Vitamin A Estimation.

Blood was obtained for vitamin A and carotene estimation by bleeding from the jugular vein with a No. 13 Birmingham Standard Wire gauge needle.

The method of analysis used for blood and liver vitamin A determination was essentially that given by the Association of Vitamin Chemists Inc. (1947).

Fifty millilitres of plasma was saponified with alcoholic potassium hydroxide, the saponified mixture extracted with petroleum ether and the extraction process repeated three times. The bulked ether extracts were washed three times with water, dried over anhydrous sodium sulphate, filtered through a No. 41 Whatman filter paper and evaporated to dryness in a stream of nitrogen. The residue was immediately taken up in moisture-free chloroform made to a convenient volume and an aliquot taken for analysis. Vitamin A was determined by the Carr-Price reaction with antimony trichloride reagent using the Lumetron photoelectric colorimeter and a 620 mμ glass filter. The instrument was calibrated by means of a U.S.P. vitamin A reference standard of crystalline vitamin A acetate in cottonseed oil contained in a special gelatin capsule. Carotene was determined on the petroleum ether extract using a 440 mμ glass filter. The reference standard was carotene (90 per cent. beta, 10 per cent. alpha) obtained from Eastman Kodak Company, Rochester, N.Y.

A similar procedure was adopted for liver analyses. Fifty grams of minced liver was weighed into a flask containing 100 ml. of 10 per cent. potassium hydroxide solution. The extraction after saponification was carried out as for blood. The petroleum ether extracts were made to 200 ml. in a volumetric flask and a convenient volume then taken for analysis as outlined above.

The results of analyses for plasma vitamin A levels were much lower than those recorded in the literature. Duplicate analyses done at the same time were satisfactory, but the results of analyses of samples taken at three-weekly intervals fluctuated widely. Some source of loss of vitamin A was suspected. This loss was ultimately traced to two causes:—

(1) Adsorption of vitamin A by the No. 41 Whatman filter paper used to filter the petroleum ether extract after drying over sodium sulphate.

(2) Adsorption of vitamin A by the sodium sulphate used as a drying agent.

The method of Peirce (1945) eliminates these two sources of error. This procedure has since been adopted and has given satisfactory results.

Because of these findings the blood vitamin A analyses have been excluded. Some loss of vitamin A also occurred in the liver analysis, but the results have been included because they still show that all rams had adequate reserves at the end of the experiment.

RESULTS.

Clinical Observations.

Weight trends in the sheep are presented in Table 1.

Group I—Basal ration.—During the seven months these sheep were in separate groups (pre-treatment and experimental periods) each animal gained between 13 lb. and 28 lb. in body weight. The lowest serum calcium estimation was 7·9 mg. per 100 ml. and blood red cell counts ranged from 6·3 million to 8 million per c.mm.

No clinical abnormalities were noticed.

Group II—Methyl-thiouracil treated.—In this group one animal died from malignant oedema (on the 45th day of the experiment) and two from hypocalcaemia (on the 29th and 51st days). A fourth animal gained only 6 lb.; its serum calcium on 50th day of experiment and at slaughter was low (6·4 mg. per 100 ml.) but the animal showed no symptoms. The remaining two rams gained 21 lb. and 30 lb. in weight; the others made comparable weight gains while not clinically affected. Red cell counts as low as 4·4 million per c.mm. were recorded at the end of the experiment. All animals were lethargic and generally inactive. This was attributed to a hypothyroid state.

Details are:—

Ram 3176.—Gained 27 lb. between July 31 and December 16. On 28th day of experiment it had a flaccid paralysis. The serum calcium was 5·9 mg. per 100 ml., and 15 g. calcium borogluconate was given intravenously. After this the beast was brighter, but on being lifted onto his feet he still could not walk normally. Inability to extend the phalanges caused conspicuous knuckling. Hypocalcaemia was suspected and the animal was killed the next day for post-mortem examination. A sample of serum taken just prior to slaughter contained 6 mg. calcium per 100 ml. and 1.95 mg. magnesium per 100 ml.

Ram 2744.—Gained 21 lb. from July 31 until death on 51st day of experiment, apparently from acute hypocalcaemia (serum calcium 5·9 mg. per 100 ml.).

Ram 2979.—Gained 21 lb. from July 31.

Ram 3021.—Gained 30 lb. from July 31.

Table 1.

LIVEWEIGHTS (IN POUNDS) AND CAUSES OF DEATH DURING THE EXPERIMENT.

Ram No.	Pre-Experimental Period.					Experimental Period.				
	31-7-50.	29-8-50.	25-9-50.	23-10-50.	10-11-50.	1-12-50.	15-12-50.	2-1-51.	23-1-51.	26-2-51.
Group I.—										
3152	96	94	94	102	103	107	103	104	107	110
3172	76	79	83	88	91	93	96	95	99	101
3004	84	91	98	101	103	100	103	105	109	110
3073	98	104	102	114	118	124	121	118	123	126
2743	89	88	91	100	104	105	103	101	105	106
3189	70	72	70	83	87	86	81	80	81	83
Group II.—										
3176	104·5	117	118	122	129	131	131*	..	..	..
2744	81	77	77	89	97	99	102	103†	..	..
2979	84	85	88	92	95	90	91	90	101	105
3021	72	75	79	82	83	88	94	90	103	101
2829	90	92	92	98	100	93	89	81	90	96
2985	91	74	100	113	112	108	95‡	..	..	..
Group III.—										
2820	74	75	67	71	69	69	66	62	62§	..
3065	81	83	89	86	96	96	97	95	102	105
2702	92	102	97	116	119	119	114	113	119	124
3163	91	94	100	103	110	110	110	106	111	114
3000	104	110	111	123	125	126	125	125	133	121
3030	101	103	103	110	116	112	110	113	117	117
Group IV.—										
2902	84	88	86	91	92	81	90	90	99	104
3026	91	95	103	103	112	108	114	115	122	117
2882	102	105	107	115	118	115	92	106	110	113
2671	102	118	119	130	131	132	136	120	132	121
2638	92	86	95	94	94	87	91	94¶	..	..
2983	83	78	84	90	100	100	74	80	88	94

* Killed on 29th day (16th December); hypocalcaemia.

† Death from hypocalcaemia on 51st day.

‡ Death from malignant oedema on 45th day.

§ Killed 93rd day; abscess of the tongue.

¶ Death from punctured kidney on 45th day.

Ram 2829.—During the first 50 days of methyl-thiouracil treatment weight dropped 20 lb.; the serum calcium was then 6·4 mg. per 100 ml. and red blood cell count 5·4 million per c.mm. During the next 50 days the weight improved 11 lb., but red blood cell count remained low at 4·4 million and serum calcium at 6·4 mg. per 100 ml.

Ram 2985.—Gained 16 lb. from July 31 up to the 21st day of experiment, when the appetite was reduced; an injured ear was swollen. On the morning of January 1, 24 days after the onset of this disease, the animal was found dead. Post-mortem examination indicated that malignant oedema was the cause of death.

Group III—Carotene supplement.—In this group only one animal (No. 2820) was clinically abnormal and was dropped from the experiment. There was no evidence of hypocalcaemia in any animal in this group.

Details are:—

Ram 2820.—Throughout the experiment did poorly; it lost 5 lb. from July 31 until treatment commenced and a further 7 lb. afterwards. During the last 50 days of the treatment period, six blood counts ranged from 3 million to 4·2 million red cells per c.mm. A serum sample taken before post-mortem examination contained 11 mg. calcium per 100 ml. Post-mortem examination on 93rd day of experiment showed a small abscess at the base of the tongue. This probably interfered with deglutition, thereby inducing malnutrition, which in turn caused anaemia. This animal will therefore be disregarded when considering the results.

The other five rams gained from 17 lb. to 32 lb. from July 31. Red cell counts ranged from 6·1 million to 8·7 million per c.mm. The lowest serum calcium was 7·2 mg. per 100 ml.

Group IV—Methyl-thiouracil treatment and carotene supplement.—In this group also, one animal (No. 2983) was dropped from the experiment on clinical grounds. Jaundice and severe anaemia developed, apparently due to dosing with methyl-thiouracil. Two other animals (Nos. 2882 and 2671) showed similar but less severe symptoms. No. 2671 was clinically hypocalcaemic. Serum calcium estimations in this group were with one exception low.

Details are:—

Ram 2902.—Gained 20 lb. from July 31. Serum calcium estimations of 7·6 mg. and 8·4 mg. per 100 ml. were recorded on 23rd and 100th days of experiment.

Ram 3026.—Gained 23 lb. from July 31. Red blood cell counts of 7·0, 7·0 and 5·4 million per c.mm. were recorded on 18th, 28th and 104th days of experiment, respectively. The calcium estimation was 4 mg. per 100 ml. on serum taken at slaughter. No symptoms of hypocalcaemia were evident.

Ram 2882.—Gained 3 lb. from July 31. Red blood count was 7 million per c.mm. on 65th day of experiment. The animal was depressed and slightly jaundiced and continued in this condition until 97th day. Dosing with methyl-thiouracil was continued during this period except on 75th and 76th days. Appetite was poor and red cell count dropped to 4 million on the 85th day. Serum calcium was 10·6 mg. per 100 ml. and the red cell count was 5 million on the day of autopsy.

Ram 2671.—Gained 33 lb. from July 31 to 32nd day of experiment. At this stage the red blood cell count was 2.5 million. On 42nd day, in addition to pale mucous membranes and slight jaundice, the beast was lame. The gait suggested tenderness of the lower limb but no injury could be detected. On 44th day the gait was stilted, the back humped and head down. A muscular tremor was obvious and the animal was lying down most of the time. Serum calcium was 5·8 mg. per 100 ml., so 15 g. of calcium borogluconate was given intravenously. No marked improvement could be seen the following day and two further doses of 8 g. and 15 g. were given. After this, gradual recovery took place. Serum calcium was 8.8 mg. per 100 ml. on 68th day. Seventeen days later stiffness and lameness were again apparent and the serum calcium was 4·2 mg. per 100 ml. Another 15 g. of calcium borogluconate was given intravenously. This effected some improvement. From then until autopsy the beast was often unable to get up in the morning, but on being lifted would eat well and appear quite bright. During this last five weeks, calcium (5 g. ground limestone every second day) was given by mouth. Just before post-mortem examination the serum calcium was 6·2 mg. per 100 ml. Five red blood cell counts made after 20th day of experiment ranged from 3 million to 4·5 million per c.mm.

Ram 2983—Fourteen days after treatment commenced, body weight was the same as at the final pre-treatment weighing 21 days earlier. On 15th day of experiment the ram was noticed to be depressed and jaundiced, and soon lost condition. At the second post-treatment weighing 13 days later it was found to have lost 26 lb. Red cell count was 1·2 million on 17th day and a week later 1·1 million. During this week the faeces became fluid, but not pale. After 10 days of sickness, an improvement in general condition was noticed. Between 17th and 28th day, treatment

with methyl-thiouracil was discontinued. It appeared that the symptoms were due to the toxicity of methyl-thiouracil. After this upset the ram was left in the group and treated similarly to the others, but was not considered to be in the experiment. By 82nd day the red cell count had returned to 3·5 million. The dose rate of methyl-thiouracil was increased to 2 g. twice a day from 83rd until 118th day and red cells were counted each week. In chronological order the red blood cell counts during this period were 3·3, 5·2, 6·2, 5·5, 5·5 million per c.mm. The effect of a higher dose rate did not confirm our suspicion about the toxicity of methyl-thiouracil. However, its apparent effect on this animal earlier, and its effect on other animals in the experiment, is evidence that it may cause jaundice and anaemia.

Ram 2638—Gained only 3 lb. from July 31 until 45th day of experiment, when the animal died from a punctured kidney. Apparently this wound was inflicted during electrical stimulation to collect semen.

Semen Value.

Analysis of semen characters showed no significant difference between the groups. The difference between the semen of rams within the groups outweighed any differences there might have been between the groups.

The semen of all rams was poor.

Vitamin A and Carotene Content.

Results of liver vitamin A analyses are recorded in Table 2. Plasma vitamin A levels have not been recorded because, as mentioned under Methods, the technique used in the earlier determinations gave inaccurate results. There was no detectable carotene in the plasma.

Post-mortem Examinations.

The essential data are presented in Table 2. (Four rams, Nos. 2743, 3189, 3000 and 3030, were not autopsied).

The conspicuous feature was the enlarged thyroid glands in all the rams treated with methyl-thiouracil. Their average weight (16·5 g.) was over four times that of thyroids from the non-treated animals (4.0 g.). Macroscopically the enlarged glands were dark burgundy in colour and fleshy in texture.

Microscopically the epithelial cells were hypertrophied, and many were over three times as high as they were wide. In some glands a little colloid was present, but in most it had almost all disappeared. In one very large (27·5 g.) thyroid from ram 3021 a considerable amount of colloid was present in large (1-2 mm.) acini (Fig. 1, a).

The glands from animals not treated with methyl-thiouracil were paler, less vascular and more fibrous. The histology was that of the classical thyroid except that cysts were present in all glands. These cysts varied in size up to

Table 2.

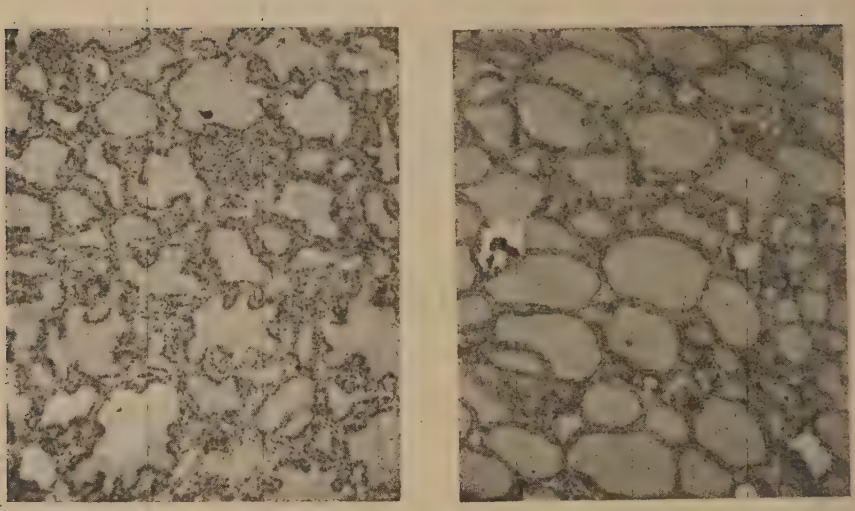
RESULTS OF ESTIMATIONS AND EXAMINATIONS.

Treatment.	Ram No.	Date of Autopsy.	Weight at Autopsy.	Weight of Thyroid.	Weight of Testes.	Serum Calcium.	Liver Vitamin A.
			Lb.	g.	g.	mg./100 ml.	micrograms/g
Pre-experiment control	3159	20–11–50	87	4	433	..	100
	3161	20–11–50	121	6	200	..	200
	2909	20–11–50	115	5·5	300	..	250
Group I.—Control	3152	22–2–51	100	4	390	8·7	65
	3172*	1–3–51	101	4	345	8·4	85
	3004	6–3–51	110	3·5	425	7·9	30
	3073	15–3–51	129	4·5	515	10·3	90
Group II.—Methyl-thiouracil	3176	16–12–50	131	11	380	6	65
	2744	7–1–51	103	23	390	5·9	..
	2979	22–2–51	105	11·6	340	6·5	110
	3021	1–3–51	102	27·5	465	4·3	165
	2829	6–3–51	96	7	285	6·4	150
Group III.—Carotene	2820†	13–2–51	61	3·5	135	11	140
	3065	22–2–51	105	4·5	445	9·5	75
	2702	1–3–51	124	3·5	525	7·2	160
	3163	6–3–51	114	4·6	450	8·9	90
Group IV.—Carotene and methyl-thiouracil	2902	22–2–51	104	13·5	360	8·4	190
	3026	1–3–51	117	16	428	4	55
	2882	6–3–51	113	19	410	10·6	85
	2671	13–3–51	125	19·6	377	6·2	95
Carotene and methyl thiouracil	2983‡	13–3–51	98	24·4	300	7·0	2

* Atrophy of one testicle.

† Abscess on base of tongue.

‡ This ram was discarded from Group IV. on December 15 because during the preceding nine days it had eaten very little and had marked jaundice and severe anaemia. Having recovered to some extent, methyl-thiouracil dosing was started again on December 17. On February 8 the dose rate was increased to 4g. per day, which was continued until post-mortem on March 15.



a *b*

Fig. 1.

Effect of Methyl-Thiouracil Treatment on Thyroid Gland.

(*a*) Thyroid gland from a ram given methyl-thiouracil. The weight of the gland was 27·5 g.

(*b*) Thyroid epithelium and colloid accumulation in a normal gland weighing 4·5 g. (*Haematoxylin and eosin*) × 80.)

about half the diameter of the gland. They were filled with pale-cream-coloured, homogeneous material of soft, cheesy consistency. Microscopically the material was faintly eosinophilic and enclosed in a thin capsule (Fig. 2). This peculiarity of the normal ovine thyroid has been mentioned by Moutaux (1940), who noticed it in two ewes.

The other constant post-mortem feature of the methyl-thiouracil treated animals was a larger liver darker in colour than those of the other rams. When the liver was minced for vitamin A sampling, this difference was very noticeable. Minced liver from the methyl-thiouracil treated rams was quite fluid and would pour, while liver from the other animals was, when minced, more viscous. Sections from all of the livers were microscopically similar.

Regarding the testes, Table 2 shows that those of methyl-thiouracil treated sheep tended to be lighter in weight.

Histological criteria for determining testicular activity have not been seen in the literature. However, it is reasonable to assume that the quantity of spermatozoa being produced would be indicated by the number of dividing spermatocytes and the presence of all stages of developing spermatozoa in the testes. Further, the degree to which the epididymis is packed with spermatozoa

should also indicate testicular activity, though this would be affected by recent ejaculation. Histological examination could not, except in extreme cases, give any indication of the quality of the spermatozoa being produced. Observations based on these criteria indicated that the testes of the methyl-thiouracil treated animals were slightly less active than those of rams not receiving methyl-thiouracil. This was probably a nutritional effect, as feed intake was depressed in the methyl-thiouracil animals. There is the possibility, however, that it was a thyroid effect, because the histological changes were similar to those described by Maqsood (1951) in rabbits given 0·1 per cent. thiouracil in their feed. This author stated that similar changes occur in rams given thiouracil.

DISCUSSION.

To minimise the carotene intake, a basal ration presumed to be of poor quality was used. The various treatments were begun after five months on the basal ration. It was anticipated that at this stage liver vitamin A reserves would be low in all groups, but the levels found in the pre-experimental controls and in Group I at the end of the experiment showed adequate vitamin A reserves, indicating that the basal ration was not deficient in vitamin A precursors.

The liver vitamin A reserves in rams from Group II and Group IV at the end of the experiment were comparable with those in the controls, thus leading to the conclusion that the hypothyroid state produced in Groups II and IV did not prevent the conversion of carotene to vitamin A.

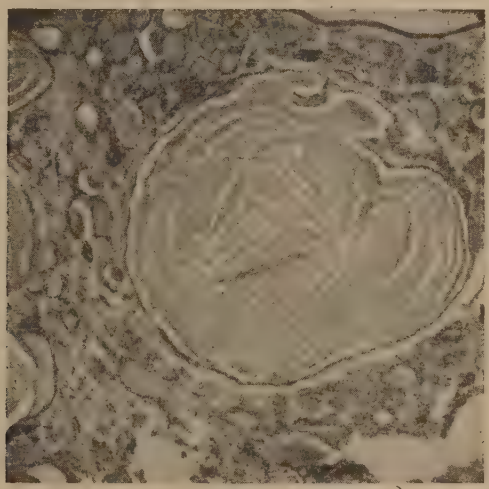


Fig. 2.

Thyroid Gland, weight 4·5 g., from an Untreated Sheep, illustrating the Nature of the Cyst commonly seen in Normal Sheep.

(*Haematoxylin and eosin;* × 80.)

The total vitamin A reserve in the treated animals was probably greater than indicated in Table 2 because the livers of these animals were enlarged.

No measurable carotene was detected in the plasma of sheep in any group. This is in agreement with observations by Eveleth, Bolin and Goldsby (1949).

No marked differences were apparent in the semen from different groups, but histological examinations indicated decreased testicular activity in the groups receiving methyl-thiouracil. This was probably caused by the lower food intake of the methyl-thiouracil treated groups.

One ram, No. 2983, after receiving 4 g. of methyl-thiouracil per day for three weeks had the very low liver vitamin A concentration of 2 micrograms per gram. This would suggest that methyl-thiouracil, possibly through the hypothyroid state resulting therefrom, affected the storage of vitamin A in the liver. However, all other rams in Groups II and III showed comparable clinical effects of hypothyroidism, but their liver vitamin A reserves were adequate

The effects of hypothyroidism on the conversion of carotene to vitamin A have been studied in rats. Johnson and Baumann (1947) and others found that the hypothyroid state was associated with lowered conversion of carotene to vitamin A. On the other hand, Heimer, Maslow and Sobel (1949) found higher liver vitamin A levels in thyroidectomised and thyroxine-treated animals kept on a vitamin A deficient diet than in controls.

That methyl-thiouracil had produced a hypothyroid state was indicated by the lethargic clinical state of the rams and by the grossly enlarged thyroids. The large, dark, flaccid livers in all of the methyl-thiouracil treated animals and the jaundice and severe anaemia in Nos. 2983, 2882 and 2671 indicate that methyl-thiouracil is to some extent toxic to sheep. This was unexpected; methyl-thiouracil is reputed to be less toxic than thiouracil, and Schultze and Turner (1945) found no toxic effects in the goat when 5 g. thiouracil per day per 100 lb. body weight was given for 30 days.

A feature that claimed our attention was the occurrence of clinical hypocalcaemia in Nos. 3176, 2744 and 2671, all of which were treated with methyl-thiouracil. All serum calcium estimations on the methyl-thiouracil treated animals were appreciably lower than in those not receiving methyl-thiouracil.

Three possible explanations of the hypocalcaemia are:—

(1) Thyroid and parathyroid glands are very closely associated in sheep. The marked hyperplasia of the thyroid may have crowded the parathyroid or may have restricted its blood supply. This could reduce the output of parathyroid hormone and thus lower the serum calcium level.

(2) An alternative is suggested by the statement of Bodansky and Bodansky (1940), after reviewing experimental evidence, that: ". . . the effect of thyroid hormone on calcium and phosphorus metabolism is essentially one of release of these elements from bone." It is therefore possible that in hypothyroid animals calcium and phosphorus would be retained in the bones and serum calcium would drop.

(3) Food intake was reduced in the methyl-thiouracil treated animals, and consequently the calcium intake would also be reduced. As poor oaten chaff is low in calcium, the reduced intake may have been below the critical level.

ACKNOWLEDGMENTS.

This work was instigated by Mr. G. R. Moule (Director of Sheep Husbandry in the Division of Animal Industry), to whom we are grateful for many suggestions. Mr. Moule and the staff of the Sheep and Wool Branch also did the semen evaluation and analysis. The Wool Research Trust Fund made possible the purchase of the animals, carotene and methyl-thiouracil used. Mr. R. K. Keith (C.S.I.R.O. Veterinary Parasitology Laboratory, Yeerongpilly) is thanked for the micro-photographs. We are indebted to Mr. A. K. Sutherland (Senior Veterinary Pathologist, Animal Health Station, Yeerongpilly) for his advice and assistance with both the experiment and the preparation of this paper.

REFERENCES.

Association of Vitamin Chemists Inc. 1947. Methods of Vitamin Assay. New York: Interscience Publishers.

Astwood, E. B., and Van Der Laan, W. P. 1945. Thiouracil derivatives of greater activity for the treatment of hyperthyroidism. J. Clin. Endocrin. 5: 424-30. (Abs. in Nutr. Abs. Rev. 16, p. 453.)

Best, C. H., and Taylor, N. B. 1945. The Physiological Basis of Medical Practice. Baltimore: Williams and Wilkins.

Bodansky, M., and Bodansky, O. 1940. Biochemistry of Disease. Toronto: The Macmillan Company.

Bogart, R., and Mayer, D. T. 1946. Environmental temperature and thyroid gland involvement in lowered fertility of rams. Mo. Agric. Expt. Sta. Res. Bull. 402.

Canadell, J. M., and Valdecasas, F. G. 1947. Action inhibitrice du thiouracil sur la carotine. Experienta 3: 35-6. (Abs. in Nutr. Abs. Rev. 17, p. 93.)

Dempsey, E. W., and Astwood, E. B. 1943. Determination of the rate of thyroid hormone secretion at various environmental temperatures. Endocrin. 32: 509.

Ely, R. E., Olson, K. J., and Reineke, E. P. 1948. Absorption and elimination of thiouracil in ruminants. J. Ani. Sci. 7: 208-13.

Eveleth, D. F., Bolin, D. W., and Goldsby, A. I. 1949. Experimental avitaminosis A in sheep. Amer. J. Vet. Res. 10: 250-4.

GUNN, R. M. C. 1936. Fertility in sheep: artificial production of seminal ejaculation and the characteristics of the spermatozoa contained therein. Counc. Sci. Industr. Res. Aust. Bull. 94.

———————, SANDERS, R. N., and GRANGER, W. 1942. Studies in fertility in sheep. 2. Seminal changes affecting fertility in rams. Counc. Sci. Industr. Res. Aust. Bull. 148.

HEIMER, C. B., MASLOW, H. L., and SOBEL, A. E. 1949. Influence of thyroid on the utilization of vitamin A. J. Nutr. 38: 345-52.

JOHNSON, R. M., and BAUMANN, C. A. 1947. The effect of thyroid on the conversion of carotene into vitamin A. J. Biol. Chem. 171: 513-21.

MAQSOOD, M. 1951. The influence of thyroid status on spermatogenesis. Science 114: 693.

MONTAUX, M. 1940. Peculiarities observées sur les thyroids de mouton. Rec. Med. Vet. 116: 17.

MOULE, G. R. 1950. Some problems of sheep breeding in semi-arid tropical Queensland. Aust. Vet. J. 26: 29-

NATIONAL RESEARCH COUNCIL (U.S.A.). 1944. Recommended nutrient allowances for domestic animals. No. 5. Recommended nutrient allowances for sheep.

PEIRCE, A. W.. 1945. The effect of intake of carotene on the general health and on the concentration of carotene and of vitamin A in the blood and liver of sheep. Aust. J. Expt. Biol. Med. Sci. 23: 295-303.

SAPSFORD, C. S. 1951. Seasonal changes in spermatogenesis in rams: their relation to plane of nutrition and to vitamin A status. Aust. J. Agric. Res. 2: 331-41.

SCHULTZE, A. B., and TURNER, C. W. 1945. The determination of the rate of thyroxine secretion by certain domestic animals. Mo. Agric. Expt. Sta. Res. Bull. 392.

THE OXALATE CONTENT OF SOME QUEENSLAND PASTURE PLANTS.

By R. H. MATHAMS, M.C., B.Sc. (formerly Analyst, Chemical Laboratory, Division of Plant Industry), and A. K. SUTHERLAND, B.V.Sc., M.S. (Senior Veterinary Pathologist, Animal Health Station, Yeerongpilly).

SUMMARY.

The water-soluble and total oxalate contents of 80 species of pasture plants were determined by K. W. Moir's modification (in press) of the method of Myers. The results are expressed as percentage of oxalic acid on a moisture-free basis.

Thirty-four species contained less than 0·5 per cent. total oxalate.

Seventeen species contained high levels of total oxalate (4·0 per cent. or more). One was a grass (Panicum antidotale); the others, all herbage (or forb) plants, were Abutilon oxycarpum, Amaranthus mitchelli, Atriplex muelleri, Atriplex semibaccata, Bassia echinopsila, Chenopodium auricomum, Chenopodium hubbardii, Chenopodium triangulare, Emex australis, Enchylaena tomentosa, Oxalis corniculata, Portulaca oleracea, Salsola kali, Tetragonia expansa, Threlkeldia proceriflora and Trianthema portulacastrum.

Seventeen grasses were examined; the only one with a significant oxalate content was Panicum antidotale with 3·5 per cent. soluble and 4·9 per cent. total oxalate.

Oxalate content is highest in immature plants, and it varies with rainfall and probably also with soil and other environmental factors.

Field and experimental evidence on poisoning of sheep and cattle by Portulaca oleracea, Salsola kali, Trianthema portulacastrum and Threlkeldia proceriflora is described and discussed. Because of their high oxalate content, these plants can cause acute poisoning, particularly in hungry or travelling stock.

INTRODUCTION.

The oxalate content of plants grazed by sheep in Queensland first attracted attention because elsewhere the toxicity of a number of species had been attributed to their high content of oxalate or oxalic acid (Fleming, 1928; Bull, 1929; Steyn, 1934). In 1948, the Department became interested in oxalate intake as one of the factors possibly associated with the occurrence of carbonate calculi in the urinary tract of sheep. The oxalate content of a number of the pasture plants grazed by sheep in Queensland was therefore determined, and the results are presented in this paper.

METHODS.

Samples of about 2 lb. of the whole of the aerial part of each plant were collected by field officers of the Department of Agriculture and Stock. Botanical identifications were made in the Queensland Herbarium. Samples were air-dried and then ground in a Wiley mill.

The method of analysis for oxalate was a modification by Moir (1953, in press) of the method described by Myers (1947).

The results are expressed as oxalic acid on a moisture-free basis. Where the total oxalate content was less than 0·5 per cent., soluble oxalate was not determined and was recorded as negative.

RESULTS.

The soluble and total oxalate contents of the 80 plant species examined, together with the localities from which the samples were obtained, are shown in Table 1.

Thirty-four species contained virtually no oxalate—i.e., less than 0·5 per cent. total oxalate.

Seventeen species contained high levels of oxalate—i.e., 4·0 per cent. or more. One was a grass (*Panicum antidotale*). The others (all herbage plants, or forbs) were:—*Abutilon oxycarpum, Amaranthus mitchellii* (boggabri), *Atriplex muelleri* (annual saltbush), *Atriplex semibaccata* (creeping saltbush), *Bassia echinopsila* (red burr), *Chenopodium auricomum* (blue bush), *Chenopodium hubbardii* (a saltbush), *Chenopodium triangulare* (a fishweed), *Emex australis* (spiny emex), *Enchylaena tomentosa* (berry cotton-bush), *Oxalis corniculata* (wood sorrel), *Portulaca oleracea* (pigweed), *Salsola kali* (soft roly-poly), *Tetragonia expansa* (New Zealand spinach), *Threlkeldia proceriflora* (soda bush), and *Trianthema portulacastrum* (black pigweed).

The marked variation in the oxalate content of samples of the same species collected in different localities and at different times is shown particularly by the results for *Portulaca oleracea* and *Salsola kali*. The figures for *Salsola kali* show also the importance of stage of growth of the plant in relation to oxalate content; in particular, a single sample was divided into two parts (plants under 6 in. and plants 6-18 inches high), which were then analysed separately with the following results:—

	Soluble Oxalate	Total Oxalate.
	%	%
Plants under 6 in.	10·7 ..	15·7
Plants 6-18 in.	9·5 ..	13·8

Variations in the oxalate content of samples of *Atriplex semibaccata* and *Tetragonia expansa* collected at approximately monthly intervals from a pasture at Goondiwindi are shown in Fig. 1, together with the monthly rainfall. In each species the young, succulent plants contained much more oxalate than the mature plants.

Table 1.

SOLUBLE AND TOTAL OXALATE CONTENT OF SAMPLES OF 80 SPECIES OF PASTURE PLANTS.
(Expressed as percentage of oxalic acid on a moisture-free basis.)

Species.	Locality and Remarks.	Oxalate.	
		Soluble.	Total.
Abutilon malvifolium	Barcaldine	*	1·5
Abutilon otocarpum	Goondiwindi	—	—
	Goondiwindi	—	1·7
Abutilon oxycarpum	Goondiwindi	—	5·9
Acacia cambagei	Barcaldine	—	—
Alternanthera nodiflora	Barcaldine	1·3	3·7
	Longreach	0·6	2·5
Amaranthus mitchellii	Barcaldine	4·6	7·2
Aristida latifolia	Barcaldine	—	—
	Winton	—	0·2
Astrebla lappacea	Winton	—	—
Astrebla (mixed species)		—	—
Atriplex muelleri	Barcaldine	4·5	6·2
	Barcaldine	4·0	7·0
Atriplex semibaccata	Goondiwindi—Dec. 1948	3·5	4·9
	Goondiwindi—Mar. 1949	3·3	4·6
	Goondiwindi—May 1949	2·3	3·8
	Goondiwindi—June 1949	2·5	3·7
	Goondiwindi—July 1949	2·5	4·0
	Goondiwindi—Aug. 1949	0·6	1·9
	Goondiwindi—Nov. 1949 (young)	7·4	8·3
	Goondiwindi—Dec. 1949	6·4	10·0
	Goondiwindi—Jan. 1950	2·8	4·1
	Goondiwindi—Sept. 1950 (young)	7·4	10·2
Basilicum polystachyon	Longreach	—	0·8
Bassia anisacanthoides	Barcaldine	0·5	5·7
Bassia echinopsila	Isisford	Trace	7·9
	Blackall	—	3·0
	Winton	0·9	2·4
	Winton	5·1	7·5
Bassia quinquecuspis	Winton	2·2	2·4
Bassia tricuspis	Isisford	—	2·5
Boerhavia diffusa	Goondiwindi	0·5	3·6
Bothriochloa ewartiana	Winton	—	—
Brachiaria purpurascens	Barcaldine	0·5	1·1
Brachycome curvicarpa	Barcaldine	—	0·5
Bulbine bulbosa	Isisford	—	—
Calotis hispidula	Isisford	—	Trace
	Winton	—	—
Cassia occidentalis	Winton	—	—
Cassinia laevis	Barcaldine	—	—
Chenopodium auricomum	Longreach	2·1	4·6
Chenopodium hubbardii	Goondiwindi—June 1949	6·3	6·5
	Goondiwindi—Nov. 1949	4·8	7·2
	Goondiwindi—Aug. 1950	4·9	8·7
	Goondiwindi—Sept. 1950	1·4	7·6
Chenopodium triangulare	Dalby	4·3	5·5
Cucumis myriocarpus (fruit)	Longreach	—	—

Table 1—continued.

SOLUBLE AND TOTAL OXALATE CONTENT OF SAMPLES OF 80 SPECIES OF PASTURE PLANTS—*continued.*

(Expressed as percentage of oxalic acid on a moisture-free basis.)

Species.	Locality and Remarks.	Oxalate.	
		Soluble.	Total.
Cucumis trigonus (fruit)	Longreach	—	—
(dry vine)	Longreach	—	—
Cyperus dactylotes	Goondiwindi	—	—
Cyperus gilesii	Barcaldine	—	—
Dactyloctenium radulans	Barcaldine	—	0·5
Daucus glochidiatus	Winton	—	0·8
	Barcaldine	—	—
	Isisford	—	2·5
Dichanthium sericeum	Barcaldine	—	—
Emex australis	Toowoomba	2·0	6·0
Enchylaena tomentosa	Dalby	6·5	6·7
	Goondiwindi	6·6	6·9
	Longreach	6·7	7·2
Enneapogon flavescens	Barcaldine	—	—
Eragrostis setifolia	Barcaldine	—	
Eremophila maculata	Longreach	—	2·1
Eriochloa pseudoacrotricha	Winton	—	—
	Barcaldine	—	—
	Goondiwindi	0·4	0·9
Euphorbia drummondii	Longreach	—	0·5
	Winton	—	—
	Barcaldine	—	
Gnaphalium japonicum	Barcaldine	—	—
Goodenia strangfordii	Winton	—	—
Iseilema membranaceum	Winton	—	—
Iseilema vaginiflorum	Barcaldine	—	—
Kochia coronata	Barcaldine	—	2·0
Lepidium hyssopifolium	Longreach	—	
Leptochloa digitata	Goondiwindi	—	—
Malvastrum spicatum	Barcaldine	*	2·5
	Barcaldine	—	0·8
	Winton	—	0·5
	Longreach	—	1·5
Minuria integerrima	Longreach	—	—
Neptunia gracilis	Barcaldine	—	0·9
Nicotiana megalosiphon	Barcaldine	—	—
Oxalis corniculata	Goondiwindi	4·1	7·0
Owenia acidula	Longreach	—	3·4
Panicum antidotale	Goondiwindi	3·5	4·9
Panicum maximum	Brisbane	0·5	1·5
Paspalidium caespitosum	Goondiwindi	0·9	1·2
Phyllanthus maderaspatensis	Longreach	—	—
	Longreach	—	—

Table 1—continued.

SOLUBLE AND TOTAL OXALATE CONTENT OF SAMPLES OF 80 SPECIES OF PASTURE PLANTS—*continued.*

(Expressed as percentage of oxalic acid on a moisture-free basis.)

Species.	Locality and Remarks.	Oxalate.	
		Soluble.	Total.
Plantago varia	Winton	—	—
	Barcaldine	—	—
	Isisford	—	—
	Goondiwindi—4 samples	—	—
Portulaca oleracea	Brisbane	*	3·5
	Dalby	4·3	5·5
	Dalby	6·8	9·0
	Millmerran	*	6·1
	Blackall	*	9·0
	Barcaldine	*	10·6
	Longreach	7·4	9·3
	Longreach	8·5	11·1
	Longreach	9·4	13·3
	Cunnamulla‡	5·1	7·9
	Cunnamulla‡	4·6	7·7
	Emerald	5·0	8·3
	Longreach	*	10·7
Psoralea cinerea	Winton	—	0·8
Pterigeron odorus	Longreach	—	0·5
Pterocaulon sphacelatum	Longreach	—	0·4
Salsola kali	Barcaldine	2·0	5·0
	Winton—Mature	4·0	10·3
	Winton—Green, 18″	4·8	6·2
	Winton—Green, 6–18″	6·4	6·6
	Longreach—Green, 12″	4·5	7·8
	Winton†—Green, less than 6″	10·7	15·7
	Winton†—Green, 6–18″	9·5	13·8
Senecio lautus	Barcaldine	—	—
Sida fibulifera	Longreach	—	2·0
	Longreach	—	1·3
	Longreach	—	—
Sida trichopoda	Barcaldine	—	1·1
	Longreach	—	—
Solanum esuriale	Barcaldine	*	2·4
	Barcaldine	—	2·8
Solanum nigrum	Winton	—	—
Sphaeranthus hirtus	Longreach	—	1·2
Tetragonia expansa	Dalby	6·6	7·1
	Goondiwindi—Dec. 1948	6·0	7·7
	Goondiwindi—May 1949	6·1	7·8
	Goondiwindi—June 1949	2·3	4·3
	Goondiwindi—July 1949	4·9	6·6
	Goondiwindi—Aug. 1949 (old)	1·5	2·9
	Goondiwindi—Aug. 1949 (young)	11·1	12·1
	Goondiwindi—Oct. 1949 (young)	11·7	13·4
	Goondiwindi—Nov. 1949	12·0	13·1
	Goondiwindi—Dec. 1949	11·2	12·1

Table 1—continued.

SOLUBLE AND TOTAL OXALATE CONTENT OF SAMPLES OF 80 SPECIES OF PASTURE PLANTS—*continued.*

(Expressed as percentage of oxalic acid on a moisture-free basis.)

Species.	Locality and Remarks.	Oxalate.	
		Soluble.	Total.
	Goondiwindi—Jan. 1950	7·7	8·9
	Goondiwindi—Aug. 1950	6·2	8·8
	Goondiwindi—Sept. 1950	6·0	11·0
Teucrium integrifolium	Longreach	..	2·0
Thelkeldia proceriflora	Barcaldine	2·0	3·6
	Barcaldine	6·2	8·6
Trianthema portulacastrum ..	Emerald	7·7	9·6
Tribulus terrestris	Barcaldine	*	0·4
Triraphis mollis	Winton	—	0·1
Verbena officinalis	Barcaldine	—	2·6
	Goondiwindi	—	—
	Longreach	—	—
Verbesina encelioides	Goondiwindi	—	0·4
Vittadenia pterochaeta	Barcaldine	—	—
Vittadenia triloba	Goondiwindi	—	0·5

* Not analysed for soluble oxalate.

† One sample from a single stand of this species was separated into two parts (plants less than 6 in. high and plants 6-18 in. high) and each part analysed separately.

‡ The Queensland Herbarium advised that these two samples from the same area in May, 1952, could be *Portulaca intraterranea* or *Portulaca oleracea* as it is not possible to distinguish these species without good flowering specimens.

Of the 17 grasses examined, only one contained a significant amount of oxalate—viz., *Panicum antidotale,* with 3·5 per cent. soluble and 4·9 per cent. total oxalate. Three species—*Brachiaria purpurascens, Panicum maximum* and *Paspalidium caespitosum*—contained from 1.0 per cent. to 1.5 per cent. total oxalate, while the following grasses contained less than 0.5 per cent. total oxalate:—*Aristida latifolia, Astrebla lappacea, Astrebla* (mixed species), *Bothriochloa ewartiana, Dactyloctenium radulans, Dichanthium sericeum, Enneapogon flavescens, Eragrostis setifolia, Eriochloa pseudoacrotricha, Iseilema membranaceum, Iseilema vaginiflorum, Leptochloa digitata,* and *Triraphis mollis.*

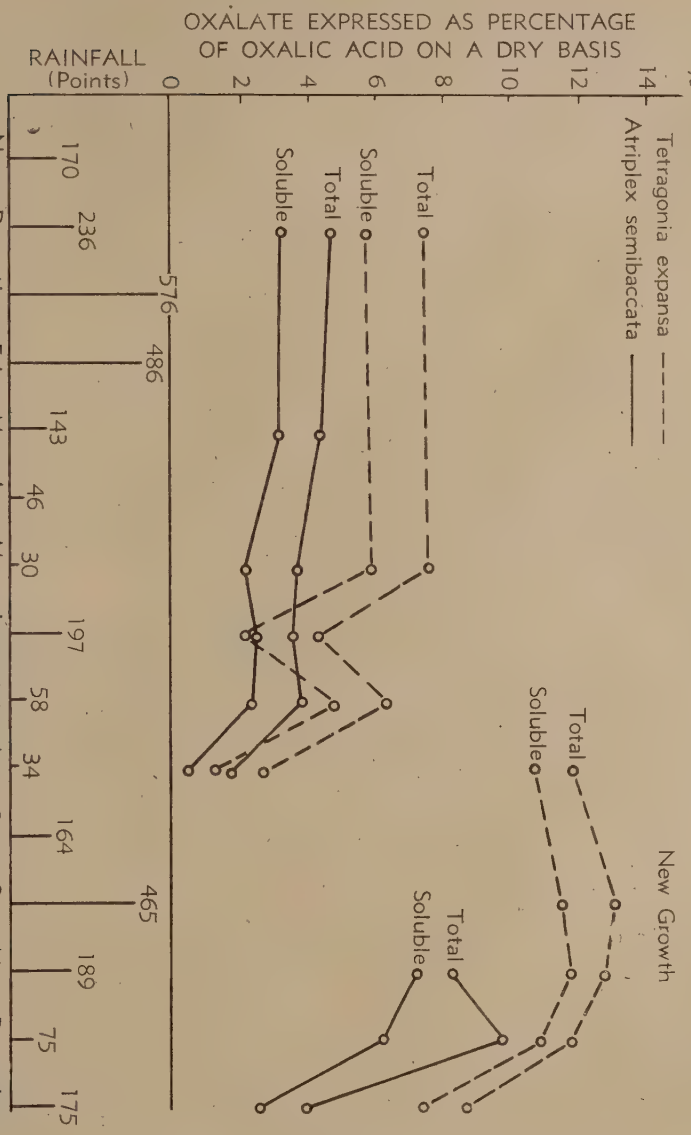


Fig. 1.

Total and Soluble Oxalate in Samples of *Tetragonia expansa* and *Atriplex semibaccata* from a Pasture at Goondiwindi. The monthly rainfall (in points) is also shown.

THE EFFECTS OF OXALATE-RICH PLANTS ON GRAZING ANIMALS.

The effects of oxalate ingestion have been summarized by Rimington and Steyn (1933) as follows:—

"(1) A local inflammation, present even when the acid is administered in dilute solution.

(2) Muscular twitching or tetany, accompanied by other nervous symptoms due to the removal of calcium ions from the system and an upset of the base balance Ca Mg/Na K.

(3) Lowered coagulability of the blood owing to the decrease in calcium ions.

(4) Lesions in the excretory organs, kidneys, etc., owing to the deposition in the cellular substance of hard, crystalline concretions of calcium oxalate."

Losses due to chronic oxalic acid poisoning were described by Bull (1929) in sheep that had grazed seven weeks or more on pastures composed almost entirely of *Oxalis cernua* in South Australia. Some sheep exhibited tetany, but the majority died as the result of extensive nephritis due to the presence in the plant of large quantities of oxalic acid.

Stafford, Franklin and Brown (1937) described acute oxalate poisoning in flocks of lactating ewes that grazed pastures heavily infested with sorrel (*Rumex acetosa*). The symptoms were those of acute hypocalcaemia. Serum inorganic calcium levels in nine affected ewes ranged from 2.45 mg. to 6.51 mg. per 100 ml., and treatment by parenteral injection of calcium gluconate was followed by rapid recovery. The trouble was attributed to the high oxalic acid content of the leaves and stems of the sorrel—up to 0.27 per cent. on green, undried material.

Hypocalcaemia and hypermagnesaemia in wethers that grazed on *Oxalis corniculata* was recorded by the New South Wales Department of Agriculture in 1938 (Hurst, 1942).

The occurrence of hypocalcaemia in sheep fed oxalate-rich plants was studied by the Australian Council for Scientific and Industrial Research (1944, 1945, 1946) and is mentioned as follows in the three Annual Reports cited:—

"Chemical analyses were made of two species of Portulaca and one of Oxalis. The values for total oxalate content ranged from 8·5 to 15·5 per cent. Consumption of these plants was possibly associated with an outbreak of clinical hypocalcaemia in ewes." (1944.)

"Force-feeding experiments on sheep with *Oxalis cernua* (soursob) and *Threlkeldia proceriflora* (soda-bush), both of which are rich in salts of oxalic acid, showed that they may cause an acute hypocalcaemia which can be cured by injection of calcium gluconate. In the case of soda-bush poisoning, a sheep thus cured of the acute symptoms developed lesions in the urinary tract." (1945.)

"A force-feeding experiment with locally grown *Oxalis cernua* (soursob) confirmed the previous finding that severe hypocalcaemia can be induced by as little as 600 grams of the undried plant, owing to its high oxalate content." (1946.)

Talapatra, Ray and Sen (1948) showed that the high level of soluble oxalate in a ration of paddy straw fed to cattle interfered with calcium assimilation.

Mortality in stock has occurred occasionally in Queensland as the result of ingestion of oxalate-rich plants—viz., *Portulaca oleracea, Threlkeldia proceriflora, Trianthema portulacastrum* and *Salsola kali*. Observations on the role of ingested oxalate in the aetiology of urinary calculi in sheep will be published in a separate paper. The following remarks are therefore confined to acute oxalate poisoning.

Portulaca oleracea (Pigweed).

This species has occasionally been suspected of poisoning sheep, but the evidence has been considered inconclusive. The fresh green plant was fed to three experimental sheep at the Animal Health Station, Yeerongpilly, in 1940 (White, Harvey and Mathams, unpublished) as follows:—

Wether No. 75, weight 65 lb., was fed at the rate of 1,400 g. per day during the first week, increasing gradually to 1,750 g. per day during the sixth and seventh weeks, after which it died; the lesions were emaciation and chronic intoxication.

Lamb No. 85, weight 44 lb., was fed 1,000 to 1,200 g. per day for 8 weeks.

Lamb No. 86, weight 49 lb., was fed 1,000 to 1,200 g. per day for 9 weeks.

The plant fed to Nos. 75 and 85 contained 6.1 per cent. total oxalate, and that fed to No. 86 contained 3.5 per cent. total oxalate (soluble oxalate was not determined). The serum calcium levels of all three sheep remained within the normal range. Lambs Nos. 85 and 86 lost some weight but showed no other ill effects, either before or after they were sacrificed for autopsy.

More recently there has been substantial field evidence that pigweed of high oxalate content can poison hungry sheep that gorge the plant.

In June 1950, deaths in a mob of 30 yearling rams were investigated at Longreach by Mr. R. B. Young (Senior Sheep and Wool Adviser). After a 24-hour journey by road and rail transport, the rams were untrucked and confined in the railway yards at 11 a.m. At 5 p.m. the same day they appeared healthy, but at 7 a.m. next morning seven were dead. The post-mortem findings (viz., acute congestion of subcutis and viscera, particularly the kidneys, and tympanites of the rumen) were consistent with acute oxalate poisoning. The bulk of the ingesta in the rumen was pigweed (stems, leaves, fruits and seeds); a much smaller amount of *Trianthema portulacastrum* (see page 328) was also present. The yards in which the rams were confined contained a great deal of pigweed, together with some harmless weeds. A sample of the pigweed sent to the laboratory contained 7.4 per cent. soluble and 9.3 per cent. total oxalate. It was concluded that death was due to acute oxalate poisoning.

A similar condition occurred in a mob of 268 rams which were examined at Cunnamulla in April 1952 by Mr. A. W. Grummitt (Inspector of Stock). The rams were unloaded after a 25-hour journey into yards containing a heavy growth of button grass and pigweed. The four affected sheep showed unsteady gait and hypersensitivity. Two died about two hours after they were first observed sick, and two others recovered after treatment with calcium borogluconate. At autopsy, enteritis and congestion of the lungs were seen. Analyses of ingesta sent to the laboratory showed the following amounts of total oxalate:—

Rumen content	2.16%
Small intestine content	2.11%

Samples of mature pigweed (after flowering) were collected subsequently from the yards. The plant was identified as probably *Portulaca oleracea*, but possibly *Portulaca intraterranea*, because specific identification is not possible in the absence of good flowering material. These samples contained oxalate as follows:—

	Soluble. %	Total. %
Sample No. 1	5·1 ..	7·9
Sample No. 2	4·6 ..	7·7

A week later 12 out of a mob of 369 mixed fat cattle died in the same yards. Inspector Grummitt reported that the cattle had been four days without feed when they were unloaded into the yards. No sick animals were examined. The one animal available for autopsy showed haemorrhages of the mucosa of the abomasum and intestine. Analysis of a sample of rumen content showed that it contained 1.7 per cent. of total oxalate.

A fourth instance of acute oxalate poisoning due to the ingestion of *Portulaca oleracea* was investigated in 1952 by Mr. R. B. Young (Senior Sheep and Wool Adviser) and is best presented by quoting from his report accompanying specimens sent to the laboratory.

"A flock of 2,100 ewes in good condition and due to lamb in four weeks were yarded for the night preparatory to crutching. Next morning they were released hungry into a paddock not used for some time and rich in pigweed and other herbage. In a few hours 100 ewes were sick, showing pallor and later cyanosis of visible mucosae, paralysis of hindquarters and bloat. They tried to rise when disturbed but soon went down again.

"Five ewes died and the remainder recovered, although it was feared at one stage that all might die. Forty ewes were treated with calcium boroguleonate, and 20 were drenched with sodium thiosulphate, but recovery occurred equally in untreated and treated sheep.

"Autopsies revealed intense congestion of the viscera. The paunches were very full and contained much gas and a great deal of pigweed."

Samples of pigweed and dried stomach content sent to the laboratory were analysed for total oxalate, with the following results:—

Portulaca oleracea	10.75%
Ingesta	4.77%

The presence in the stomach of much pigweed and a high level of oxalate is considered to confirm the field diagnosis of acute oxalate poisoning due to the ingestion of pigweed.

Salsola kali (Soft Roly Poly).

This annual or perennial herb is abundant in western Queensland. The mature plant is dry and easily broken, so it is carried by the wind and becomes piled against fences and other obstacles. Although the mature plant is quite unattractive to stock, the immature stage is heavily grazed by sheep and cattle.

Zellner (1927) found that *Salsola kali* is rich in potassium salts and also contains appreciable oxalic acid. Hurst (1942) stated that the plant had been suspected of causing mortality in cattle in New South Wales, but no particulars were given. McClymont (1947) listed it as one of the common weeds in New South Wales which are rich in oxalates.

The plant is suspected to have caused mortality in stock on several occasions in Queensland, shown in the records and reports of officers of the Department as follows:—

1939—In pigs and calves at Chinchilla.

1940—In hungry cattle at Hughenden.

1942—The loss of 800 out of several thousands of sheep that travelled on stock routes in the Julia Creek district was attributed to this plant.

1947—Mortality in cattle at Pittsworth was attributed to young *Salsola kali* three to five inches high, which was heavily grazed. The cattle had just been untrucked after travelling from a dry area.

The toxicity of *Salsola kali* was tested at the Animal Health Station, Oonoonba, by Mr. R. E. Churchward (Veterinary Officer) in 1940. The following particulars are from his unpublished report.

One pound of plant (stage of growth not stated) was extracted with one litre of water for 24 hours; the fluid was then expressed and administered by stomach tube to sheep weighing 80-90 lb. The results were:—

SHEEP No. 19.

1st day—extract from 1 lb. given

2nd day—extract from 1 lb. given

7th day—extract from 2 lb. given

8th day—found dead.

Autopsy revealed intense congestion of the mucosa of the abomasum and duodenum. The kidneys were soft with petechiae in the cortex, and the capsule stripped easily. Subcutaneous blood vessels were engorged, and the blood throughout the body was dark and not completely clotted.

SHEEP No. 16.

1st day—ate 1 lb of freshly chaffed *Salsola kali*.

2id day—aqueous extract from 2 lb. of plant.

3rd day—refused freshly chopped plant.

4th day—refused to eat fresh plant; appeared normal; discharged.

SHEEP No. 18.

1st day—aqueous extract of 2 lb. *Salsola kali*.

2nd day—aqueous extract of 2 lb. *Salsola kali*.

3rd day—apparently normal.

4th day—appeared normal; discharged.

It would appear that at the time of these observations the possibility of the toxicity of *Salsola kali* being due to its oxalate content was not considered. The findings on sheep No. 19 are consistent with death from acute oxalate poisoning. It is evident, too, from the results shown in Table 1 that the amount of soluble oxalate in immature *Salsola kali* (4—10 per cent.) would be likely to cause acute oxalate poisoning, particularly when heavily grazed by hungry stock.

Trianthema portulacastrum (Black Pigweed).

The loss of four of a group of 30 fat cattle at Emerald was investigated in February 1952, by Mr. J. T. Littleton (Inspector of Stock). The cattle were yarded for about three days and then grazed on a heavy stand of *Trianthema portulacastrum* and some *Portulaca oleracea*. Symptoms exhibited were diarrhoea, dyspnoea and salivation. The gait was short and the head was held forward with tongue protruded. Later the faeces were scanty. The animals died in a sleeping position. Autopsy revealed inflammation of the abomasum and duodenum. Pigweed was present in the rumen.

Samples of the two pigweeds obtained from the area contained oxalate as follows:—

	Soluble. %	Total. %
T. portulacastrum	7·7	9·6
P. oleracea	5·0	8·3

Threlkeldia proceriflora (Soda Bush).

Heavy stands of this weed occur at times in central-western Queensland, particularly on overgrazed areas such as stock routes and reserves. Field and experimental evidence on its toxicity to sheep was reported by Legg and Francis (1939). They described mortalities that occurred when hungry sheep were allowed to graze heavy stands of the plant. They found that 1 lb. of fresh plant killed a wether which had been starved for 24 hours. The force-feeding experiments mentioned earlier (Australia, Council for Scientific and Industrial Research, 1946) indicate that the toxicity of soda bush is due to its oxalate content.

Instances of poisoning by soda-bush have not been investigated by us. The oxalate content of one of the samples examined was rather low (3.6 per cent. total), but the second was quite high (6.2 per cent. soluble and 8.6 per cent. total) and would be dangerous to hungry stock that gorged on it.

Bassia echinopsila (Red Burr).

This is one of the common winter herbage species of central-western Queensland. It is extensively grazed by sheep wherever it occurs. Only four samples have been examined for oxalate (Table 1). One sample (from Winton in September 1952) consisted of plants 1½-6 in. high at the early fruiting stage and contained 5.1 per cent. soluble and 7.5 per cent. total oxalate. Another sample contained 7.9 per cent. total oxalate but only a trace of soluble oxalate. The remaining two samples were older plants and did not contain appreciable oxalate.

The only record of *B. echinopsila* having been suspected of causing poisoning in stock is a report by Mr. M. N. S. Jackson (Senior Sheep and Wool Adviser)*. In a mob of 2,000 lambing ewes 50 died in January 1948 when they were held for 24 hours prior to crutching in a paddock in the Winton district containing a heavy stand of red burr. The symptoms and post-mortem findings were consistent with acute oxalate poisoning, and were in fact noted as being similar to those of poisoning with *Threlkeldia proceriflora*. At the time of these field observations it was not appreciated that red burr may at times contain a high level of soluble oxalate.

Tetragonia expansa (New Zealand Spinach).

It is shown in Fig. 1 that samples of this species range from as low as 2.9 per cent. total oxalate in mature plants to the very high figure of 12.0 per cent. soluble and 13.1 per cent. total oxalate in immature plants. However, there is no record of its having come under suspicion as a cause of mortality in sheep or cattle in Queensland. This is probably due to the fact that it is palatable to sheep only when it is mature and rather dry (S. L. Everist, personal communication, 1952).

* We are indebted to Mr. W. D. Francis, Government Botanist, for drawing our attention to this record.

DISCUSSION.

Among the 17 grasses examined, *Panicum antidotale* was the only one with a significant oxalate content (3·5 per cent. soluble and 4·9 per cent. total). Talapatra, Ray, Kehar and Sen (1942) listed nine grass species in which they found only small quantities of oxalic acid, and in addition they reported the following as containing significant amounts:—

	Anhydrous Oxalic Acid (Dry Basis).	
	Total.	Soluble.
	%	%
Paddy straw—		
No. 1	1·46	1·25
No. 2	1·60	1·35
Napier grass (*Pennisetum purpureum*)—		
Early cut	3·05	2·04
Dead ripe	0·65	0·40
Guinea grass (*Panicum maximum*)—		
Early cut	2·00	1·10
Dead ripe	0·80	0·25

They considered these levels of oxalic acid to be high, particularly in plants in the early stages of growth. The amounts are higher than those in most of the grasses we examined, but they are much lower than the amounts in some of the common Queensland herbage (forb) species—e.g., the saltbushes (*Atriplex muelleri* and *Atriplex semibaccata*), *Chenopodium hubbardii*, *Portulaca oleracea*, *Salsola kali*, and *Trianthema portulacastrum*.

Talapatra, Ray and Sen (1948) showed in India that the high level of oxalate in a ration of paddy straw interfered with the calcium assimilation of cattle. They showed further that the soluble oxalate was the fraction that interfered with calcium metabolism, while the insoluble fraction (probably in the form of calcium oxalate) was excreted unchanged in the faeces. It would seem, therefore, that the toxicity of an oxalate-rich plant is due chiefly to its content of soluble oxalate.

The experiments of Fleming (1928) with *Sarcobatus vermiculatus* (greasewood) in Nevada, U.S.A. provide valuable information on the amounts of plant and the circumstances required to cause acute oxalate poisoning in sheep. The analyses reported on six samples of the plant showed 11·40 per cent. to 15·39 per cent. anhydrous oxalic acid on a moisture-free basis, but the method used would be likely to give figures 2–3 per cent. too high. It is therefore probable that the plant Fleming used for feeding experiments contained about 10 per cent. total oxalate. (The soluble oxalate content was not reported).

The results of 36 feeding tests with leaves and green stems of greasewood, in which the amounts of plant shown were force-fed to sheep in 30–60 minutes, are summarized as follows:

Result.	Number of Feeding Tests.	Amount of Plant Fed in Lb. per 100 Lb. Liveweight.
Death	36	4·50–8·69 (mean 5·61)
Sickness and recovery	9	3·53–6·11 (mean 5·06)
No symptoms	11	1·87–5·02 (mean 3·88)

The smallest dose that caused symptoms was 3·53 lb. per 100 lb. liveweight, but in three tests more than 4·00 lb. and in two tests more than 5·00 lb. failed to cause any symptoms. Fleming remarked that this showed that "the individuality of the animal and its condition influence the degree to which it is subject to this form of poisoning." He did not mention that variations in oxalate content of different samples of plant might also be important. It can be assumed, however, that the fatal dose of greasewood is about 5·00 lb. per 100 lb. liveweight when it is consumed within an hour.

Two ewes were fed greasewood mixed with the daily ration of grasses, clovers and weeds at the rate of 4·00 and 5·00 lb. per 100 lb. liveweight, and no ill effects were produced. In a third series of experiments, greasewood equivalent to 5·0, 6·3, 5·88 and 6·69 lb. per 100 lb. was fed in two, nine, three and three force-feedings during the day (9 a.m. to 5 p.m.) No ill effects were produced. Fleming therefore concluded that "sheep become poisoned by greasewood as a result of eating the leaves and green stems almost exclusively in a quantity approximating the stomach capacity during a relatively short time."

These experiments are consistent with the observations recorded above in connection with poisoning by *Portulaca oleracea*, *Salsola kali*, *Trianthema portulacastrum* and *Threlkeldia proceriflora*—viz., that travelling or hungry stock are particularly apt to be affected. Such stock often eat in a short time large quantities of plants that local stock eat in only small quantities. Further, acute poisoning may be apt to occur in hungry stock because of more rapid absorption of oxalate.

The natural pastures in western Queensland usually contain a great variety of plant species; so, except in special circumstances, such as those encountered by travelling stock, a large quantity of any single species is not likely to be eaten by grazing animals. It has been frequently observed also that stock accustomed to pastures containing oxalate-rich plants do not suffer acute poisoning—perhaps their grazing habits are such that large amounts of oxalate are not ingested at one time. The instances of *Oxalis cernua* poisoning described by Bull (1929) in South Australia and of sorrel poisoning described

by Stafford, Franklin and Brown (1937) in New Zealand occurred in sheep grazing heavy, almost pure, stands of these weeds which had developed on cultivation paddocks.

The most acute manifestation of poisoning by oxalate-rich plants would appear to be a poisoning by oxalate *per se*, while in the less severe manifestation hypocalcaemia or alkalosis may be the prominent feature. In animals whose calcium metabolism is delicately balanced, such as the lactating ewes described by Stafford, Franklin and Brown (1937), hypocalcaemia is especially evident.* In many field outbreaks both manifestations occur. Recovery from acute oxalate poisoning entails the risk of subsequent symptoms attributable to chronic lesions in the urinary tract (Australia. Council for Scientific and Industrial Research, 1946).

Little information is available on the toxic doses of oxalic acid and its salts for sheep and cattle. New Zealand workers (New Zealand Department of Agriculture, Animal Research Division, 1952) have reported on the toxicity of sodium oxalate for cattle. They dosed four cows by mouth with 100 g., 200 g., 400 g. and 600 g., respectively, in five equal doses in two hours. The results were:—

100 g.—No effect.

200 g.—Symptoms which were alleviated by three injections of calcium within 10 hours.

400 g. and 600 g.—Died in convulsions within four hours of the first drench.

In the three affected cows, the most marked feature was the rapid drop in serum calcium.

According to Frohner (1919), a sheep (weight 35 kg.) that received 25 g. of oxalic acid—i.e. 0·7 g. per kg.—in water (concentration not mentioned) died in four hours.

M. White, J. M. Harvey and R. H. Mathams (1940, unpublished) examined the toxicity for sheep of sodium oxalate given by mouth in 2 per cent. aqueous solution. Their results (Table 2) show that one lamb given 1 g. sodium oxalate per kg. body weight died of acute oxalate poisoning. One lamb showed no ill effects from a dose of 0.5 g./kg. per day for 23 weeks, while another lamb given 0·37 g./kg. per day died in the 22nd week of the experiment.

* Field observations indicate that clinical hypocalcaemia (which responds to calcium therapy) often occurs in western Queensland when pregnant or lactating ewes are mustered and confined for 24-48 hours for operations such as crutching or shearing. The trouble is more prevalent during good winter seasons when herbage is abundant (K. J. Astill, personal communication, 1952). In addition to exercise and fasting, which have been shown experimentally to precipitate clinical hypocalcaemia in ewes (Franklin, 1948a, 1948b), heavy feeding on oxalate-rich herbages might also play a part in these outbreaks.

Table 2.

TOXICITY OF SODIUM OXALATE GIVEN ORALLY IN 2% AQUEOUS SOLUTION TO SHEEP 50—60 LB. LIVEWEIGHT.

(After White, Harvey and Mathams, 1940, unpublished.)

Lamb No.	Dose Rate per Day g./kg.	Duration of Treatment.	Result.
135	1·0	Once only	Died less than 24 hours
88	1·0	2 days	Died
90	1·06	6 days	Killed—moribund
89	1·0	11 days	Died
140	0·5*	23 weeks	Negative
141	0·37*	22 weeks	Died
142	0·32*	23 weeks	Negative
143	0·28*	23 weeks	Negative

* Divided into four doses each day.

It is evident that the figures given in Table 1 are a general guide only to the oxalate content of a plant. Oxalate content is highest in immature plants, and it varies also with rainfall, and probably also with soil and other environmental factors. Diurnal periodicity in the oxalic acid content of the leaves of *Oxalis cernua* and many desert succulent plants has been mentioned by Clarke (1934). We have noticed, too, that when plant samples have been dried slowly, the oxalate content has usually been lower than would be expected, but we have no exact data on this point. Again, we have observed that specimens that reached us in a mildewed condition have shown lower amounts of oxalate, particularly soluble oxalate, suggesting that some of it may be destroyed by fungi.

There is also another difficulty that arises in attempting to assess the oxalate intake of sheep. As the plant matures and becomes lignified, the soluble oxalate content decreases, so analysis of the entire plant probably does not give a true indication of the oxalate consumed by an animal that usually selects the young, more tender parts of the plant.

ACKNOWLEDGMENTS.

We are deeply indebted to Mr. K. W. Moir, B.Sc. (Analyst) and Mr. J. M. Harvey, M.Sc. (Senior Chemist) for analysing many of the samples reported. Mr. A. Canty (Inspector of Stock) kindly collected samples in the Goondiwindi district over a long period. Specimens and reports used in compiling the manuscript have been provided also by Messrs. R. B. Young and G. R. Morrison (Senior Sheep and Wool Advisers), Mr. J. F. Meehan (Inspector of Stock), the staff of the Queensland Herbarium and many other officers of the Department.

REFERENCES.

AUSTRALIA. COUNCIL FOR SCIENTIFIC AND INDUSTRIAL RESEARCH. 1944. Seventeenth Annual Report of the Council for Scientific and Industrial Research for the year ended 30th June, 1943. (p. 22.)

———. 1945. Nineteenth Annual Report of the Council for Scientific and Industrial Research for the year ended 30th June, 1945. (p. 36.)

———. 1946. Twentieth Annual Report of the Council for Scientific and Industrial Research for the year ended 30th June, 1946. (p. 27.)

BULL, L. B. 1929. Poisoning of sheep by soursobs (*Oxalis cernua*): chronic oxalic acid poisoning. Aust. Vet. J. 5: 60-9.

CLARKE, G. H. 1936. Important weeds of South Australia. No. 9.—Soursob (*Oxalis cernua*, Thunb.). J. Dept. Agric. S. Aust. 38: 481-505. (Reprinted in S. Aust. Dept. Agric. Bull. 313, 1936.)

FLEMING, C. E. 1928. The greasewood (*Sarcobatus verminulatus*), a range plant poisonous to sheep. Nev. Agric. Expt. Sta. Bull. 115.

FRANKLIN, M. C. 1948a. The influence of fasting or a falling plane of nutrition on the serum-calcium level. *In* Counc. Sci. Industr. Res., Aust. Bull. 240.

———. 1948b. The influence of exercise on the serum-calcium level. *In* Counc. Sci. Industr. Res., Aust. Bull. 240.

FROHNER, E. 1919. Lehrbuch der Toxicologie. Stuttgart: Verlag von Ferdinand Enke. (Cited by Steyn, 1934.)

HURST, E. (compiler). 1942. The Poison Plants of New South Wales. Sydney: New South Wales Poison Plants Committee.

LEGG, J., and FRANCIS, W. D. 1939. *Threlkeldia procriflora*. A plant poisonous to stock. Aust. Vet. J. 15: 168-71.

MCCLYMONT, G. L. 1947. Diseases of breeding ewes. Hypocalcaemia and pregnancy toxaemia. N.S.W. Dept. Agric. Diseases of Animals Leafl. 52.

MYERS, A. T. 1947. Seasonal changes in total and soluble oxalates in leaf blades and petioles of rhubarb. J. Agric. Res. 74: 33-47.

MOIR, K. W. 1953. The Determination of Oxalic Acid in Plants. Qld. J. Agric. Sci. (In press.)

NEW ZEALAND DEPARTMENT OF AGRICULTURE, ANIMAL RESEARCH DIVISION. 1952. Annual Report, Animal Research Division of the New Zealand Department of Agriculture, 1950-51.

RIMINGTON, C., and STEYN, D. G. 1933. *Psilocaulon absimile* N.E. Br. as a stock poison. I. Determination of oxalic, malic, tartaric acids, etc. Onderstepoort J. Vet. Sci. 1: 439-55.

STAFFORD, J., FRANKLIN, M. C., and BROWN, I. M. 1937. Sorrel poisoning in ewes. Canterbury Agric. Coll. Res. Rpts., Mar. 1937.

STEYN, D. G. 1934. The Toxicology of Plants in South Africa. Johannesburg: Central News Agency.

TALAPATRA, S. K., RAY, S. C., KEHAR, N. D., and SEN, K. C. 1942. Oxalic acid in feeding stuffs and its significance in calcium assimilation. Science and Culture 8: 209-13.

———, and SEN, K. C. 1948. Calcium assimilation in ruminants on oxalate-rich diet. J. Agric. Sci. 38: 163-75.

ZELLNER, J. 1927. Zur chemie der halophyten. Monatsch. Chem. 47: 611-8.

THREE NEW SPECIES OF DACINAE (TRYPETIDAE DIPTERA) FROM QUEENSLAND.

By A. W. S. MAY, M.Agr.Sc., Entomologist, Science Branch, Division of Plant Industry.

SUMMARY.

Three new species—Strumeta alyxiae, S. notatagena and Afrodacus tigrinus—are described.

INTRODUCTION.

Recent studies concerned with recording native hosts and the seasonal behaviour of certain species of the sub-family Dacinae have revealed three new species among material identified from North Queensland. Their descriptions are presented here so that the names can be incorporated in a paper dealing with hosts of the Dacinae occurring in this State, which will appear in the next issue of this Journal.

Strumeta alyxiae n.sp.

A small fly with characteristic infuscation on the wings, clear costal cells, large facial spots pointed anteriorly and the greater part of the thorax and abdomen dark brown to black. The female has a dark brown apical spot on the upper surface of each femur.

Female:—Length, excluding ovipositor, 5·6 mm.; wing 4·8 mm.

Male:—Length 5·7 mm.; wing 4·8 mm.

Head:—*Frons* fulvous, length 1·3 times width; orbital spots and anteromedial blotch tending dark brown; bristles: 1 *s. or.*, 2 *i. or.* *Lunule* shining fulvous. *Antennae* slightly longer than face, fulvous, except for outer surfaces of third segment, brown; third segment more than three times second, latter with weak pale dorsal bristle; arista long, dark brown except basal portion fulvous. *Face* pale fulvous, shiny, large subcircular facial spot, pointed anteriorly, in each antennal groove. *Palps* fulvous, flattened. *Genae* pale fulvous with pale brown subocular spot; bristle weak, black. *Occiput* fulvous, occipital row weak, approximately five on each side. *Vertex* fulvous, may be dark brown along transverse carina; ocellar triangle black; bristles: ocellar present, very weak; post-verticals strong, dark brown to black.

Thorax:—Largely dark brown to black except for following areas which are pale reddish brown: medial portion of mesonotum; below and behind postsutural stripes; a narrow band bordering humeral and notopleural calli; immediately behind outer portion of suture and beneath humeral calli. *Mesonotum* between paired shiny longitudinal black vittae covered with hoary pubescence. Also with the following yellow markings: humeral and notopleural calli; pair of postsutural stripes narrowing posteriorly and ending just before

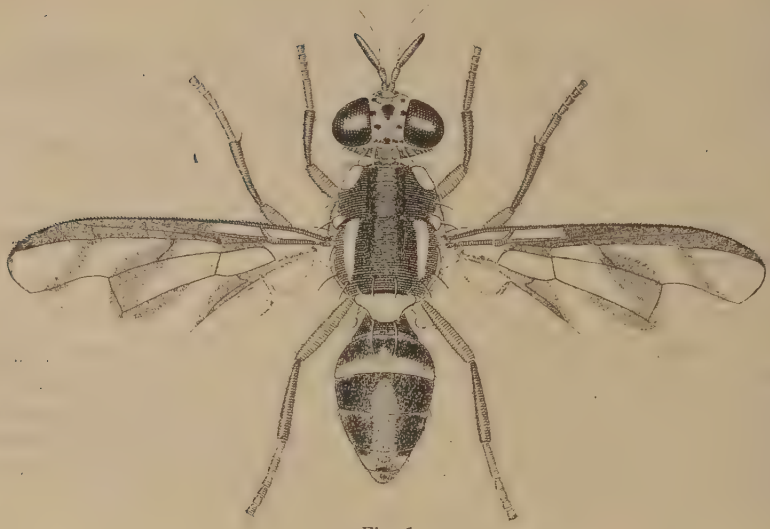


Fig. 1.

Strumeta alyxiae ♂

upper *p.sa.* bristles; a triangular mesopleural stripe, with anterior edge slightly concave, commencing at posterior margin of humeral callus and ending on sternopleuron; ¾ of upper and ⅔ of lower hypopleurals. (Note:—Newly emerged individuals are more usually pale reddish brown with the following dark brown markings which later coalesce to give the general dark brown colour of aged individuals: paired bands on mesonotum extending from anterior border to point slightly more than midway between suture and posterior margin; bordering inner margin of each postsutural stripe; before suture and bounded by each longitudinal band, the notopleural calli and hind margin of humeri; on suture behind notopleural calli; beneath lower *p.sa.* bristles; bordering anterior margin of mesopleural stripe; greater portion of sternopleuron; at base of wing and in front of lower hypopleurals; above hind coxae; basal ⅓ of lower and ½ of upper hypopleurals.) *Postnotum* brown except for lateral margins broadly dark brown to black. *Scutellum* yellow with dark brown basal band. *Halteres* pale yellowish. *Bristles*: *scp.* 4 (middle pair weak), *npl.* 2, *mpl.* 1, *a.sa.* 1, *p.sa.* 2, *prsc.* 2, *sc.* 2 (apical), *pt.* 1 (weak), all dark brown. *Legs*: coxae dark brown, remainder fulvous except for tibiae dark brown, and in females a dark brown apical spot on upper surface of femora; apical spur on middle tibiae. *Wings*: Costal cells hyaline, the second cell shorter than stigma, the proportion being 0·67. Costal band fuscous including stigma, cell R_1, whole of cell R_{2+3} and extending slightly beyond end of vein r_{4+5}. A narrow fuscous transverse band commencing on r_{4+5}, midway between junction with *r-m* and wing margin, and running transversely to join m_{1+2} midway between its junction with *Im* and the wing

margin. A second, U-shaped, fuscous band enclosing *r-m* and widening as it extends obliquely backwards to reach *Im*, then turning at inner margin of wing to run as a thinner band below and for approximately three-quarters of the length of the discal cell. Above median cell fuscous. Anal streak fuscous, not reaching wing margin; *r-m* cross vein oblique and somewhat sinuate, the section of vein m_{1+2} before and after it being in the proportion of 1·2; cell Cu restricted basally in female but almost parallel-sided in male, its length in proportion, to $cu_1 + 1a$ being 2·0 in male and 1·4 in female. Supernumerary lobe poorly developed in males. Microtrichia covering wing except for first costal cell, lower $\frac{2}{3}$ of second costal cell, median cell, cell R except in that portion above median cell, basal $\frac{1}{4}$ of discal cell except for thin diagonal band and immediately below discoidal cross vein.

Abdomen:—*Male*: dark brown to black except for following: a transverse pale fulvous band between first and second tergites, the posterior third of second tergite whitish except for central pale fulvous area, a fulvous medial band on third and fourth tergites, widening to occupy practically the whole of the fifth tergite. Shining fulvous spots on fifth and a row of cilia on post-lateral margin of third tergite. No medial stripe. *Female*: colouration as for male except that shining spots may be darker brown, and fulvous areas on third, fourth and fifth tergites are less clearly defined. A dark brown median stripe may occur on the fifth tergite in some specimens. *Ovipositor* broad, fulvous, flattened.

Holotype ♂, allotype ♀, 6 ♂ ♂ and 5 ♀ ♀ paratypes, bred from fruits of *Alyxia spicata* R.Br., Cook Highway, near Mossman, Queensland, August, 1951. (A. W. S. May).

Types in Queensland Museum; holotype Reg. No. T. 5275, allotype Reg. No. T. 5276.

This species somewhat resembles *S. recurrens* Hering in general colouration and size but differs in having no well-defined medial stripe on the abdomen and a dissimilar pattern on the wing. Both *S. frenchi* (Frogg.) and *S. umbrosa* (Fabr.) also have two transverse bands on the wing but can be differentiated from *S. alyxiae* by their size, the position of the *r-m* cross vein and the markings of the abdomen.

Strumeta notatagena n.sp.

A medium-sized fly with a distinct black spot on each cheek, elongate oval facial spots, a broad costal band, brown costal cells and a strongly developed though short ovipositor.

Female:—Length, excluding ovipositor, 5·9 mm.; wing 5·1 mm.

Head:—*Frons* fulvous, flattened, length 1·2 times width; orbital spots and medial blotch faintly brown; 1 *s.or.*, 2 *i.or.* bristles. *Lunule* discoloured though darkened. *Antennae* slightly longer than face, fulvous except for outer surface and extremity of third segment fuscous; third segment almost three times

Fig. 2.

Strumeta notatagena. ♀

second. *Face* shiny fulvous, except area between antennal grooves whitish (this may be discoloured); a well defined oval black spot occupying more than $\frac{1}{3}$ length of antennal groove. *Genae* fulvous with large black subocular spot. *Occiput* centrally brown, margin fulvous; occipital row wanting. *Vertex* fulvous; ocellar bristles weak; postverticals strong, black.

Thorax:—Mesonotum reddish-brown with longitudinal median black band as wide as outer scapulars anteriorly but broadening slightly at posterior margin to almost attain width of scutellum, and interrupted with reddish brown areas in region of suture. Pleurae mostly dark brown to black except reddish brown beneath humeri and immediately beneath wing. Yellow markings as follows: humeral callus except anterior margin brown; notopleural callus; broad parallel-sided postsutural lateral stripes reaching to upper *p.sa.* bristle; narrowly triangular mesopleural stripe ending on sternopleuron, with anterior edge straight and commencing beneath anterior notopleural bristle; $\frac{3}{4}$ of lower and $\frac{2}{6}$ of upper hypopleurals. *Postnotum* centrally brown, marginally dark brown. *Scutellum* yellow, with very narrow dark basal band. *a.sa.* bristle weak, reclinate; all other bristles strongly developed, black. *Legs* fulvous except hind tibiae, middle and fore femora brown. Coxae dark brown. *Wings* hyaline except fuscous costal band and anal streak. Costal band occupying stigma, cell R_1, outer portion of cell R_{2+3} and ending just beyond extremity of vein r_{4+5}; the remainder of cell R_{2+3} brown. Costal cells brown, with microtrichia over all but basal $\frac{1}{3}$ of first cell. Almost whole of vein

cu pale tending fulvous. *r-m* cross vein oblique, straight, the section of vein m_{1+2} bordering discal cell, before and after it being in the proportion of 1·4. Anal cell extension restricted basally, its length in proportion to $cu_1 + 1a$ being 1·4.

Abdomen:—Not broad. Dark brown to black except yellowish brown band along junction of first and second tergites; whitish on posterior half of second; paired yellowish-brown spots centrally placed on posterior margins of third and fourth tergites. Yellowish-brown around inner margins and posterior to large brown shining spots on fifth tergite. Sixth tergite pale fulvous. Basal segment of ovipositor strongly developed, convex, broad based, dark brown, shiny. Inversion membrane with four equally placed longitudinal brown ridges.

Holotype ♀, *ex* lure traps in mangoes, Kamerunga Experiment Station, Cairns, Queensland, 1952. (A. W. S. May.)

Type in Queensland Museum, Reg. No. T. 5277.

Though resembling *S. fuscatus* Perk. & May in general colouration of thorax and abdomen, these two species can be separated readily on the wing markings. The coloured costal cells and abdominal pattern separate *S. notatagena* from both *S. cacuminata* Hering and *S. bilineata* Perk. & May, two species with black medial markings on the thorax. *S. bryoniae* (Tryon), though agreeing somewhat in having a broad costal band, is a much larger species with an entirely black thorax and different abdominal markings.

AFRODACUS Bez.

The species described below has been placed in this genus provisionally but its correct status can only be decided when a male has been examined. The combination of characters, namely, 2 *sc.*, 2 *pr. sc.*, no *a.sa.* bristles, conform to this genus.

Afrodacus tigrinus n.sp.

A relatively small but robust fly with characteristic yellowish brown and black markings on the abdomen, a yellow oval medial blotch towards the posterior margin of the mesonotum, short, thick antennae, and no facial spots.

Female:—Length, excluding ovipositor, 5·3 mm.; wing 4·5 mm.

Head:—*Frons* fulvous, broad, parallel-sided, length 1·3 times width, antero-medial blotch yellowish; bristles: 1 *s.or.*, 2 *i.or.* *Lunule* shining fulvous. *Antennae* not as long as face, thickened, uniformly coloured yellow-brown; arista brown, very long. *Face* shiny, pale fulvous; no facial spots. *Palps* flattened, fulvous. *Genae* pale, no subocular spot evident; bristle weak, black. *Occiput* broadly fulvous on margins, medially brown to dark brown; occipital row wanting. *Vertex* fulvous; ocellar triangle black; bristles: ocellar present, very weak; postverticals strong, black.

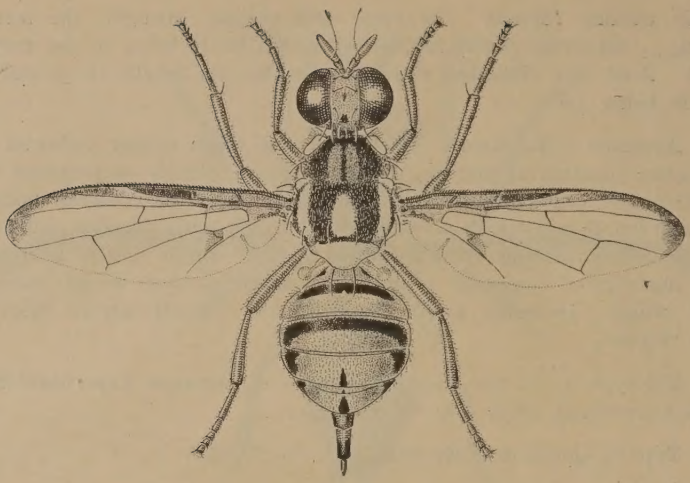


Fig. 3.
Afrodacus tigrinus. ♀

Thorax:—Broad, punctulate, black. A hoary pubescence covering medial portion of mesonotum, and with following markings:—brown: beneath humeri, above second coxae, between humeral and notopleural calli, below and behind postsutural stripes; yellow: humeral calli; broad parallel-sided postsutural stripes extending to and including upper *p.sa.* bristles; an oval postsutural medial blotch, pointed anteriorly, widening to touch the *prsc.* bristles laterally and ending at posterior margin of mesonotum; notopleural calli; a broad triangular mesopleural stripe with base attaining humeral callus and apex ending on upper sternopleuron; $\frac{3}{4}$ of upper and $\frac{2}{3}$ of lower hypopleural calli. *Scutellum* yellow with black basal band. *Postnotum* black. *Bristles*: *scp.* 4 (brown, of approximately equal length), *npl.* 2 (strong, black), *mpl.* 1 (black), *p.sa.* 2 (lower black, upper brown to dark brown), *prsc.* 2, *sc.* 2 (apical, brown), *pt.* 1 (weak). *Legs* fulvous; apical spur on middle tibiae. *Wings* hyaline. Costal cells fulvous (microtrichia covering whole of second and upper $\frac{1}{3}$ of basal cell), the second cell shorter than stigma, the proportion being 0·7 Costal band fuscous, occupying stigma, most of cell R_1, the extremity of cell R_{2+3} and broadening into an apical spot between the extremities of veins r_{4+5} and m_{1+2}; a pale area on costal band at the extremity of vein r_1. Base of cell R_{2+3} and also towards its apex beneath costal band, together with basal portion of cell R, fulvous. *r-m* oblique, sinuate, the proportion of vein m_{1+2} before and after it being in the proportion of 1·4. Anal streak pale fulvous, short. Extension of cell Cu short, parallel-sided, its length in proportion to $cu_1 + 1a$ being 1·7. Microtrichia covering wing except over basal $\frac{2}{3}$ of first costal cell, median cell, a small portion of cell R beneath junction of stigma with second costal cell, the extension of cell Cu and the alula.

Abdomen:—Tergites yellowish tending fulvous except for following clearly defined black markings:—spots on anterior margins of first; band across anterior ¼ of second; a band along anterior portion of third, which is continued laterally and terminates on fifth. A distinct medial black stripe commencing at posterior margin of fifth but fading and narrowing before attaining anterior margin only to appear again on distal half of fourth. Shining spots not conspicuous. Sixth tergite pale creamy yellow. Sternites pale. Ovipositor strongly developed, shining fulvous, broad at base, surface convex.

Holotype ♀; 1 ♀ paratype *ex* lure traps in mangoes, Kamerunga Experiment Station, Cairns, Queensland, December, 1951, and February, 1952. (A. W. S. May).

Type in Queensland Museum, Reg. No. T. 5274.

This species somewhat resembles *A. javanensis* Perk. in general colouration but differs in the relative position of the medial postsutural stripe, the absence of a well-defined abdominal stripe, the markings on the face and the lack of infuscation on the posterior cross vein.

52
T. P.

INDEX.

★ ★

PAGE.

Aberdeen, J. E. C.—
" Investigations on the phytotoxicity of Bordeaux mixture to tomatoes " .. 1

Benzene hexachloride in soil 41

Bordeaux mixture, phytotoxicity to tomatoes 1

Climatological studies of Queensland 185

Dacinae, new species 335

Drought studies 185

Everist, S. L. (with G. R. Moule)—
" Studies in the environment of Queensland. 2. The climatic factor in drought " 185

Fluorosis of sheep 47

Fodder plants—
Nutritive value 169
Oxalate content 317

Hall, W. T. K. (with J. M. Harvey)—
" Effects of methyl-thiouracil on liver vitamin A, spermatogenesis and calcium metabolism of Merino rams " 301

Harvey, J. M.—
" Chronic endemic fluorosis of sheep in Queensland " 47
" The nutritive value of some Queensland fodders " 169

Harvey, J. M. (with W. T. K. Hall)—
" Effects of methyl-thiouracil on liver vitamin A, spermatogenesis and calcium metabolism of Merino rams " 301

Mathams, R. H. (with A. K. Sutherland)—
" The oxalate content of some Queensland pasture plants " 317

May, A. W. S.—
" Potato tuber moth (*Gnorimoschema operculella* (Zell.)) investigations in southern Queensland " 142
" Three new species of *Dacinae* (Trypetidae, Diptera) from Queensland " .. 335

McDougall, W. A.—
" A note on pot experiments with ' Gammexane ' in soil " 41

Methyl-thiouracil, effects on rams 301

Moule, G. R. (with S. L. Everist)—
" Studies in the environment of Queensland. 2. The climatic factor in drought " 185

Oxalate content of pasture plants 317

Potato tuber moth investigations 142

Sheep—
Chronic endemic fluorosis 47
Effects of methyl-thiouracil 301

Sutherland, A. K. (with R. H. Mathams)—
" The oxalate content of some Queensland pasture plants " 317

Tomatoes, phytotoxicity of Bordeaux mixture 1

A. H. Tucker, Government Printer, Brisbane.